# Comparison of Aquatic Macroinvertebrate Samples Collected Using Different Field Methods

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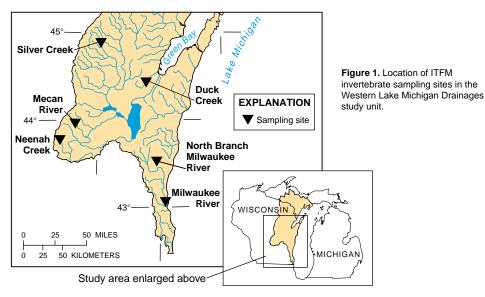
#### INTRODUCTION

Government agencies, academic institutions, and volunteer monitoring groups in the State of Wisconsin collect aquatic macroinvertebrate data to assess water quality. Sampling methods differ among agencies, reflecting the differences in the sampling objectives of each agency. Lack of information about data comparability impedes data sharing among agencies, which can result in duplicated sampling efforts or the underutilization of available information. To address these concerns, comparisons were made of macroinvertebrate samples collected from wadeable streams in Wisconsin by personnel from the U.S. Geological Survey-National Water Quality Assessment Program (USGS-NAWOA), the Wisconsin Department of Natural Resources (WDNR), the U.S. Department of Agriculture-Forest Service (USDA-FS), and volunteers from the Water Action Volunteer-Water Quality Monitoring Program (WAV). This project was part of the Intergovernmental Task Force on Monitoring Water Quality (ITFM) Wisconsin Water

# Intergovernmental Task Force on Monitoring Water Quality (ITFM)

The Intergovernmental Task Force on Monitoring Water Quality is a national program intended to develop an intergovernmental framework for water-quality monitoring to: 1) coordinate monitoring programs; 2) evaluate existing data collection activities; 3) identify the roles of Federal, State and local entities; 4) address the use of environmental indicators and standard descriptors of aquatic conditions for measuring status and trends; and 5) develop a nationwide water-information network.

A pilot ITFM project, the Wisconsin Water Resources Coordination Project, was established to coordinate and integrate waterquality monitoring. The objectives of the Project were: 1) to identify common monitoring objectives, coordinate station selection and monitoring activities, and initiate the use of common information systems; 2) to evaluate field and laboratory methods and quality control/quality assurance and to compare the various agencies water-quality collection methods for the end purpose of data sharing and promoting the use of comparable methods; and 3) to promote the development and standardization of data-analysis and datareporting techniques.



Resources Coordination Project. The numbers, types, and environmental tolerances of the organisms collected were analyzed to determine if the four different field methods that were used by the different agencies and volunteer groups provide comparable results. Additionally, this study compared the results of samples taken from different locations and habitats within the same streams.

#### SAMPLING METHODS

Sampling sites on six streams of varying size, type, and water quality in the Western Lake Michigan Drainages–NAWQA study unit were selected for sampling to ensure that different macro-invertebrate communities were sampled. The Milwaukee River in Milwaukee County, the North Branch of the Milwaukee River in Sheboygan County, Duck Creek in Brown County, Silver Creek in Shawano County, the Mecan River in Waushara County, and Neenah Creek in Adams County were sampled (fig. 1, table 1). The streams were sampled

on three days in May 1995. Sampling was coordinated among agencies to avoid sampling the same spot twice. Areas that were immediately downstream of bridges, near impoundments and stream margins or areas that contained large amounts of silt or aquatic vegetation were avoided. The sampling locations were approached from downstream to minimize disturbance of the sampling location, and samples were collected in a downstream-toupstream order to avoid including dislodged and drifting organisms from previous sampling. All agency samples were preserved using non-denatured ethanol and analyzed by the same laboratory with the exception of the samples collected by WAV which were identified in the field. Visual surveys of watershed quality and riparian and instream habitat were completed independently by each agency. Collectors qualitatively categorized the importance of factors they observed that affect water quality as being: not present, insignificant, or significant.

#### USGS-NAWQA Method

The USGS–NAWQA samples were collected from stream riffles employing a 60 centimeter deep, 425-micron net on a 50-by-33 centimeter rectangular frame called a Slack3 sampler (a modified surber sampler developed by Keith Slack, USGS, Menlo Park, Calif.) that is placed downstream of a 0.5-by-0.5 meter sampling area. All fist size or larger rocks lying 50 percent or more within the sampling area were held in front of the net, and

**Table 1.** Physical characteristics of streams sampled as part of the ITFM invertebrate sampling comparison [Ag, agricultural; I, industrial; U, urban; Rec, recreational; P, pristine; warm, warm water; cold, cold water].

Stream	Description	Stream Order	Drainage Area (mi²)		
Milwaukee River	Ag, I, U, warm	5	696		
Duck Creek	Ag, warm	4	95.5		
North Branch Milwaukee River	Ag, warm	3	51.4		
Mecan River	Rec, cold	2	28.5		
Neenah Creek	Rec, cold	2	24.6		
Silver Creek	P, cold	2	15.8		

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Collecting a macroinvertebrate kick sample from riffle substrate (USGS-NAWQA method).

scrubbed with a brush to dislodge attached organisms. The sampling area was then disturbed by digging into the substrate to a depth of 0.1 m. Additional organisms were collected by kick sampling (standing in the sampling area and shuffling the feet to dislodge macroinvertebrates from substrate which, along with some sediments and debris, drift with the current into the net) for 30 seconds (Cuffney and others, 1993). This process was repeated at three locations within the same riffle, and the samples were composited. Two such composited samples were collected, one at a downstream riffle and a second at an upstream riffle. The composite samples often contained an unacceptably large volume of material (more than 0.75 liters). In such cases, larger rocks, debris, and vegetation in the sample were discarded after ensuring that all attached organisms had been removed from the debris.

Inorganic sediments were removed from the sample by placing it in a five gallon bucket half filled with water. The sample contents were stirred by hand to suspend as much material as possible. The bucket was then swirled and decanted onto a 425-micron mesh screen until the sediment front reached the lip of the bucket. The process was repeated until it appeared that only sand and gravel remained in the bucket. These sediments were then examined for macroinvertebrates before the sediments were discarded, particularly for case-building caddisflies and small mollusks which are heavy

Collecting a macroinvertebrate snag sample from an instream snag (USDA-FS method).

and not easily removed with this method. Samples were placed in 1-liter sample jars and then preserved for processing at the laboratory. The USGS collects additional macroinvertebrate data as part of the NAWQA program which includes a qualitative sample collected from every available habitat in a reach of a determined length and a sample from a depositional area. However, these samples were not collected as part of this sampling comparison.

#### **WDNR Method**

The WDNR sample locations were in riffles with flow velocities of 0.3 meters per second or greater that contained substrate which consisted of coarse gravel to medium-sized cobble/rubble whenever possible. The WDNR collects macroinvertebrates from snag habitats when adequate riffles are not available. However, in this study all WDNR samples were collected from riffles. The WDNR's field procedure manual does not specify net dimension and requires the net to have a Stan-

dard U.S. No. 30 or finer mesh size. A 25-by-46 centimeter, rectangular net with a mesh size of 589-microns (Standard U.S. No. 30) is commonly used and was used by the WDNR collector for this comparison. The net was placed in the stream riffle, and the substrate was disturbed by kicking in the area immediately upstream of the net until it was obvious that over 125 arthropods had been collected (about 2 minutes). This sampling area was larger or smaller depending on the collector and the abundance of macro-invertebrates in each stream.

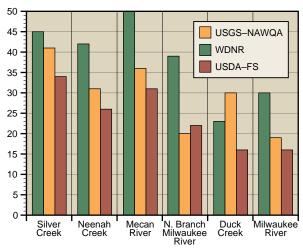
The sample was washed in the stream to remove fine sediments. Large debris was washed of macroinvertebrates in the net, and then the debris was discarded. All remaining material was placed in the sample jar

for processing at the laboratory. Three replicate samples were collected at each site by moving from downstream to upstream. The three replicate samples were processed individually, and the mean of water-quality measures for the three samples was calculated and was used to derive a single water-quality value for the site.

# USDA-FS Method

The USDA-FS collected macroinvertebrates using a 1400 micron mesh D-frame net that measured 30 centimeters along the bottom and 22 centimeters tall at the widest point. Three samples

were collected from each site. The first sample was collected from a riffle substrate and a second sample was collected from a snag habitat (overhanging grasses, weeds, trees, logs, etc.). The riffle samples were collected by placing a net on the stream bottom and kicking an area immediately upstream of the net. Individual rocks were picked up, and attached macroinvertebrates were removed from them and placed in the sample. Snags were sampled by scraping them with the net or by shaking overhanging trees and grass directly over the net. Sampling was performed until a target number of 125 organisms were collected or, in areas of low macroinvertebrate abundance, until the person or persons sampling had been collecting for a total of 1.5 combined person hours. The third sample was collected from the habitat that contained the greatest number of organisms during the first two samplings. The Forest Service uses this method because many streams in the National Forests in Wisconsin lack coarse substrate or riffles and to ensure that the macroinvertebrate sample is representative of all habitat types. Macroinvertebrates were picked from the debris collected in the net and were counted in the field. To ensure complete taxa coverage, pieces of grass and small woody debris which potentially contained invertebrates were often included in the sample. Each sample was placed in a separate 1-quart sample jar and then preserved for processing at the laboratory.



**Figure 2.** Number of taxa collected by each agency as part of the ITFM invertebrate sampling comparison.

# **WAV Method**

The WAV used kick samples to collect macroinvertebrates from a riffle at only one stream; Duck Creek. One person held two D-frame nets while another person kicked the substrate above the net for 3 minutes at each of two locations near the upstream side of the riffle. After each sample in the net was washed and large debris was removed, the samples were placed in a sample pan. Collectors then picked the macroinvertebrates, counted them, and identified to order as many of them as possible using an illustrated key in the field.

## Laboratory Methods

Macroinvertebrate sample processing, enumeration, and taxonomic identification for samples from each of the agencies were done by the Benthic Macroinvertebrate Laboratory at the University of Wisconsin—Stevens Point. Every organism collected was not always identified, rather each sample was evenly distributed in a sorting tray marked with 5-by-5 centimeter numbered grids (total 15 grids). A grid square was selected using a random numbers table and all organisms in the selected square were identified and counted. Organisms within subsequent sequentially numbered grid squares were identified until 125 organisms or more were identified at the completion of a grid square. Organisms were identified to the lowest

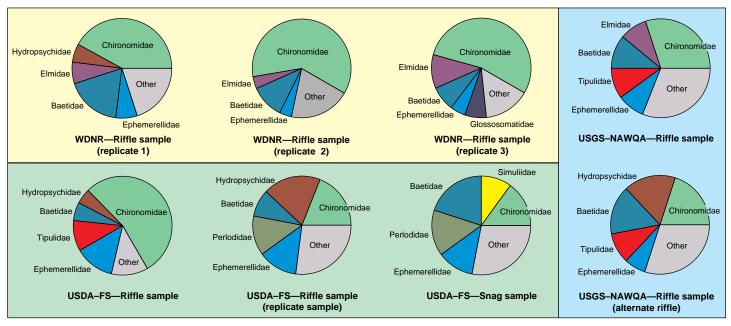


Figure 3. Community structure of invertebrate samples collected by three agencies from the Mecan River as part of the ITFM invertebrate sampling comparison.

level possible (generally species level), using recently published regional and global keys. Only identifiable specimens with assigned tolerance values were used to calculate the biotic indices and community measures. The percentage of the sample that was identified was calculated by dividing the number of grids from which organisms were identified by the total number of 15 grids. Enumeration measures used the number of taxa identified and the percentage of the total sample those identified taxa comprised to estimate the total number of individuals collected.

# **RESULTS**

# Macroinvertebrate Communities

The total number of taxa in the richest replicate sample collected by each agency from each stream is shown in figure 2. The WDNR collected the greatest number of taxa from five of the six streams. The USDA-FS generally collected the fewest taxa. Species and genera richness also followed these trends (table 2) which may be related to the methods used and the microhabitats sampled. The WDNR method generally sampled a larger area and may have encompassed more microhabitats resulting in a greater number of taxa collected. The USDA-FS method may have had a low capture efficiency of dislodged organisms that allowed smaller macroinvertebrates to pass through the larger mesh USDA-FS net. These smaller organisms were collected by the other agencies using finer meshed nets. These trends may only apply to this study because WDNR and USDA-FS use varying net mesh and sample area sizes and a target number for collection while the USGS-NAWOA uses a standard net mesh and sample area size.

Macroinvertebrate sampling methods can significantly affect the taxa collected because a particular sampling method may more effectively collect organisms from one type of habitat than another. The community structures indicated by samples examined in this study were most similar between WDNR replicate samples collected from the same riffle. These samples tended to contain very similar proportions of the same macro-invertebrate taxa. The USGS replicate substrate samples

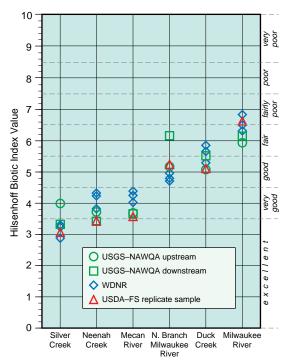
collected from two different riffles in one stream reach contained varying populations or proportions, or both, at one half of the streams sampled. No apparent changes in actual water quality were evident in the reach, therefore sample differences may be attributed to varying amounts and types of habitat at each riffle. The most taxonomically dissimilar samples tended to be those collected from snags by the USDA-FS. Samples from every stream except Neenah Creek showed the macroinvertebrate communities of snags contained several different taxa than communities collected from bottom substrate in riffles. Sampling methods determined which habitats were sampled and, therefore, affected the macroinvertebrate community structure found, even when samples were collected from the same stream reach. The samples collected at the Mecan River typify these taxonomic trends and differences found (fig. 3).

For all agencies, macroinvertebrate samples collected from the same riffle at each stream contained similar taxa, but the samples tended to contain varying proportions of individual taxa. This suggests that specific sampling methods preferentially collected certain types of macro-invertebrates. The USGS method of digging deeper into and scrubbing the substrate appears to have increased the proportion of some taxa (such as Chironomidae, which live deeper in the substrate; or Simuliidae, which attach themselves firmly to the substrate). Several of the USDA-FS samples were dominated by a particular taxa that did not dominate in samples collected by other agencies from that stream. This may be attributed to the sampling method used by the USDA-FS, which does not limit the collector to one location, but rather focuses on obtaining 125 or more organisms. The collector's effort to reach the 125 organism sample size may cause the collector to target certain microhabitats abundant with particular macroinvertebrates while bypassing areas containing other, less abundant macroinvertebrates. The larger mesh size net may have missed small macroinvertebrates and caused the

macro-invertebrates collected to appear more abundant than they were. The total number of macroinvertebrates collected using the USDA–FS method was always less than the number collected by the other agencies because the USDA–FS picked 125 macroinvertebrates in the field while the other agencies field processed the entire sample. This field picking process may also bias samples because larger and more visible organisms may be chosen while less visible or rarer organisms are excluded.

## Interpretation of Water Quality

Macroinvertebrate samples collected by each of the three agencies interpreted water-quality conditions similarly for all six streams using Hilsenhoff's Biotic Index (HBI) (Hilsenhoff, 1987) (fig. 4). The HBI is an estimate of water quality based on the



**Figure 4.** HBI values of samples collected by each agency from six streams in Wisconsin as part of the ITFM macroinvertebrate sampling comparison.

**Table 2.** Values of macroinvertebrate measures that indicate water quality, and indices that would typically be used by agencies participating in the ITFM invertebrate comparison sampling (USGS-NAWQA—mean of two replicate samples from an upstream and a downstream riffle; WDNR—mean of three replicate samples from same riffle; USDA-FS—value from replicate sample).

Stream and agency		нві	FBI	Mean tolerance value	% EPT genera	Species richness	Genera richness	Margalef's diversity index	Ratio of scrapers to collectors
Milwaukee River:	USGS-NAWQA	6.04	6.20	6.52	13	22	20	3.24	9
The same of the sa	WDNR	6.64	6.56	6.05	19	28	24	3.64	12
CONTRACTOR OF THE PARTY OF	USDA-FS	6.63	6.83	6.75	6	18	17	3.83	0
Duck Creek:	USGS-NAWQA	5.29	6.30	5.18	27	21	18	2.94	24
	WDNR	5.61	5.71	5.45	40	20	16	3.02	24
	USDA-FS	5.10	4.92	5.27	45	12	11	1.89	13
North Branch	USGS-NAWQA	5.66	6.32	5.59	24	26	23	3.22	20
Milwaukee River:	WDNR	4.83	4.80	4.97	41	32	25	3.78	36
	USDA-FS	5.31	4.95	4.89	59	19	17	3.25	50
Mecan River:	USGS-NAWQA	3.67	4.02	3.72	36	35	28	4.12	24
	WDNR	4.22	4.72	3.73	32	33	27	3.09	24
	USDA-FS	3.56	3.95	4.04	52	29	21	3.74	8
Neenah Creek:	USGS-NAWQA	3.56	4.34	3.65	30	25	16	2.92	14
The same of the sa	WDNR	4.13	4.49	3.85	27	34	24	3.42	14
	USDA-FS	3.43	4.00	3.75	31	21	13	3.15	11
Silver Creek:	USGS-NAWQA	3.65	4.12	3.49	38	39	32	4.24	14
A STATE OF THE PARTY OF	WDNR	3.14	3.33	3.33	45	38	33	3.79	21
	USDA-FS	3.06	4.16	2.97	50	32	24	3.50	17

tolerance of aquatic macroinvertebrates to organic pollution and associated reductions in dissolvedoxygen concentrations. HBI values range from 1.5 to 2 units within a single water-quality rating category. The HBI value for each sample at every stream ranged within 1 unit of the median HBI value for all samples at that respective stream; less variability than a single water-quality rating category. The variations of HBI values among the agencies' samples were the same as the variations of HBI values in replicate samples collected by a single agency. No sampling method consistently interpreted a higher or lower water-quality rating based on the HBI. All these factors indicate that each agency's methods interpreted water quality similarly for each stream and suggest that the HBI is a robust measure of water quality that is not differentially influenced by the three collection methods.

Study findings indicate that the different field collection and processing methods used resulted in assessments of different habitats, and collection of different total numbers and proportions of individual taxa. However, water quality ratings given by indices based on environmental tolerance values were similar among agencies for the macroinvertebrate taxa that were collected.

Trends similar to those indicated by the HBI were indicated by Hilsenhoff's Family Level Biotic Index (FBI) (Hilsenhoff, 1988) and by the mean tolerance value measure (Lillie and Schlesser, 1994). The HBI was positively correlated with FBI and mean tolerance value measures (r2 = 0.86 and 0.91 respectively). The WAV which performed an in-field, water-quality rating of Duck Creek, gave it a "good" rating, which is the same rating assigned to it by the government agency's samples. Other measures such as the percentage EPT (Ephemeroptera, Plecoptera, and Trichoptera), Margalef's diversity index (MDI), and several trophic function measures had significant variability between the agency's samples. However, no trends could be seen which indicate that this variability was a function of sampling methods.

The visual, qualitative watershed survey results

showed that qualitative habitat and physical setting categorizations were not consistent among the agencies. The bias of the collectors as well as differences in on-site observations and previous knowledge of the sites all seemed to affect the categorization of each stream by the different groups. These qualitative surveys were not sufficient to interpret the influence of physical setting or habitat on macroinvertebrate community measures.

### **SUMMARY**

The sampling methods used by each agency in this study tended to assess the macroinvertebrate community structure found in each stream differently. These differences may be attributed to differences in the habitat sampled by each method. Sharing of macro-invertebrate data may not be feasible when information on specific species as-

semblages is required. However, differences in community structure did not affect the ability of measures based on environmental tolerance values (HBI, FBI, and Mean Tolerance Value) to rate

water quality similarly. Information about other macroinvertebrate measures (table 2) was inconclusive because of the variability encountered in these measures. This study was unable to determine if this variability was caused by differences in sampling method or was inherent to these measures.

This study examined only part of the routine macroinvertebrate sampling done by the agencies. This study shows that differing riffle sample collection methods effect macroinvertebrate sampling results. Field collection methods need to be considered when comparing macroinvertebrate data among agencies. Comparisons of macroinvertebrate data collected using methods other than those described in this report may not produce the same results as this study.

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